

**S/N Unknown**

**PATENT**

**IN THE UNITED STATES PATENT AND TRADEMARK OFFICE**

Applicant:	John H.J. Petrini et al.	Examiner:	Unknown
Serial No.:	Unknown	Group Art Unit:	Unknown
Filed:	Herewith	Docket:	800.019US3
Title:	METHODS TO ALTER LEVELS OF A DNA REPAIR PROTEIN (as amended)		

**PRELIMINARY AMENDMENT**

Commissioner for Patents  
Washington, D.C. 20231

Sir:

**In the Specification**

On page 1, before "Statement of Government Rights", please insert

**--Cross-Reference to Related Applications**

This application is a divisional of U.S. application No. 09/067,641 filed April 27, 1998.--

Please substitute page 11, paragraph 4 for the paragraph in the appendix entitled "Clean Version of Page 11, Paragraph 4". Specific amendments to page 11, paragraph 4 are detailed in the following marked-up paragraph:

Figure 6. Structure of the p95 cDNA. (A) The schematic diagram represents the structure of the p95 cDNA. The entire 4,483 basepair (bp) cDNA [(SEQ ID NO:1)] is represented by the thin line and the rectangular box is the 754 amino acid (aa) open reading frame (ORF) (SEQ ID NO:2). Within the ORF the grey box indicates the N-terminal region showing homology to *S. cerevisiae* Xrs2. The solid line above the ORF indicates the region cloned by two-hybrid screen utilizing hMre11 as bait. (B) N-terminal alignment of p95 (SEQ ID NO:3) with Xrs2 (SEQ ID NO:4). The shaded boxes indicate the regions of similarity. The two proteins show 28% identity and 46% similarity over the region displayed. The following amino acids were considered similar: {D, E, N, Q} {F, W, Y} {I, L, V} {K, R} {A, G} {S, T} {C} {H} {M} {P}. (C) A Zoo-Blot Southern blot (Clontech, Palo Alto, CA) of EcoRI digested DNA from various species was probed with the *NBS1* cDNA. Lane 1, human; lane 2, monkey; lane 3, rat; lane 4, mouse; lane 5, dog; lane 6, cow; lane 7, rabbit; lane 8, chicken; and lane 9, yeast. The position of size markers (in kilobase pairs) is indicated on the left.

Please substitute page 13, paragraph 5 for the paragraph in the appendix entitled "Clean Version of Page 13, Paragraph 5". Specific amendments to page 13, paragraph 5 are detailed in the following marked-up paragraph:

Figure 14. cDNA sequence of p95 (SEQ ID NO:1).

Please substitute page 73, Table 1 for the table 1 in the appendix entitled "Clean Version of Page 73, Table 1". Specific amendments to page 73, table 1 are detailed in the following marked-up table:

Table 1

## Peptides Obtained From Mass Spectrometry Analysis

Peptide <sup>a</sup>	Position <sup>b</sup>
-QPPQIESFYPPPLDEPSIGSK-	189-209 (SEQ ID NO:9)
-LSSAVVFGGGGEAR-	238-251 (SEQ ID NO: 10)
-WIQSIMDMLQR-	289-299 (SEQ ID NO: 11)
-QGLRPIPEAEIGLA VIFMTTK-	300-320 (SEQ ID NO: 12)
-TTTPGPSLSQGVSVDEK-	335-351 (SEQ ID NO: 13)
-MLSQDAPTVKE-	395-404 (SEQ ID NO: 14)
-TSSNNNSMVSNTLAK-	409-423 (SEQ ID NO: 15)
-IPNYQLSPTKLPSINK-	426-441 (SEQ ID NO: 16)
-NYFQPSTKK-	458-465 (SEQ ID NO:17)
-NKEQHLSSENPVDTNSDNNLFTDTDLK-	503-529 (SEQ ID NO:18)
-EMDDVAIEDEVLEQLFK-	552-558 (SEQ ID NO: 19)
-MDIETNDTFSDEAVPESSK-	595-613 (SEQ ID NO:20)
-ELKEDSWAK-	625-635 (SEQ ID NO: 21)
-KLLLTEFR-	653-660 (SEQ ID NO:22)
-NPSGINDDYGQLK- <sup>c</sup>	671-683 (SEQ ID NO:23)
-EESLADDLFR-	736-745 (SEQ ID NO:24)

**In the Title**

Please amend the title from "DNA ENCODING A DNA REPAIR PROTEIN", to --  
METHODS TO ALTER LEVELS OF A DNA REPAIR PROTEIN--.

**In the Claims**

Please substitute the claim set entitled "Clean Version of Pending Claims" attached hereto for the pending set of claims. Specific amendments to individual claims are detailed below.

Please cancel claims 1-4, 7-15 and 19 without prejudice.

Please amend the claims as follows:

5. (Amended) A method of altering the amount of a DNA repair polypeptide in a cell, comprising:
  - (a) introducing into a host cell [the] an isolated nucleic acid molecule comprising a nucleic acid segment encoding a vertebrate DNA repair polypeptide having a molecular weight of about 95000 Da as determined by SDS-PAGE, or a biologically active fragment thereof, [of claim 1] operably linked to a promoter functional in the host cell, so as to yield a transformed host cell; and
  - (b) expressing the nucleic acid molecule in the transformed host cell as recombinant DNA repair polypeptide, wherein the amount of the recombinant polypeptide produced by the transformed cell is different than the amount of the DNA repair polypeptide produced by a corresponding untransformed cell.
6. (Amended) A method of altering the amount of a DNA repair polypeptide in a cell, comprising:
  - (a) introducing into a host cell a DNA segment comprising the complement of at least a portion of [the] a nucleic acid molecule [of claim 1] comprising a nucleic acid segment encoding a vertebrate DNA repair polypeptide having a molecular weight of about 95000 Da as determined by SDS-PAGE, or a biologically active

fragment thereof, operably linked to a promoter functional in the host cell, so as to yield a transformed host cell; and

- (b) expressing the DNA segment in the transformed host cell as antisense RNA so as to decrease the amount of the DNA repair polypeptide in the transformed cell.

16. (Amended) A transgenic mouse whose cells contain a chimeric DNA sequence, said chimeric DNA sequence comprising:

a transcription control sequence and [the] an isolated nucleic acid molecule [of claim 1] comprising a nucleic acid segment encoding a vertebrate DNA repair polypeptide having a molecular weight of about 95000 Da as determined by SDS-PAGE, or a biologically active fragment thereof, wherein the transcription control sequence and the nucleic acid molecule are operatively linked to each other and are integrated into the genome of the mouse, and wherein the nucleic acid molecule is expressed in the transgenic mouse so as to result in said mouse exhibiting increased amounts of the DNA repair polypeptide.

17. (Amended) A method of using a transgenic mouse to screen for an agent that modulates a DNA repair polypeptide, comprising:

- (a) administering the agent to the transgenic mouse, wherein the transgenic mouse comprises a chimeric DNA sequence comprising a transcription control sequence operatively linked to [the] a nucleic acid molecule [of claim 1] comprising a nucleic acid segment encoding a vertebrate DNA repair polypeptide having a molecular weight of about 95000 Da as determined by SDS-PAGE, or a biologically active fragment thereof, wherein the chimeric DNA sequence is integrated into the genome of the mouse, and wherein the nucleic acid molecule is expressed as the DNA repair polypeptide in the transgenic mouse; and
- (b) determining whether said agent modulates the amount of the DNA repair polypeptide in the transgenic mouse relative to a transgenic mouse of step (a) which has not been administered the agent.

Please add the following new claims:

20. (New) The method of claim 5, 6 or 17 wherein the nucleic acid segment comprises SEQ ID NO:1.
21. (New) The method of claim 5, 6 or 17 wherein the nucleic acid segment encodes SEQ ID NO:2.
22. (New) The method of claim 5 or 6 wherein the host cell is a mammalian host cell.
23. (New) The mouse of claim 16 wherein the nucleic acid segment comprises SEQ ID NO:1.
24. (New) The mouse of claim 16 wherein the nucleic acid segment encodes SEQ ID NO:2.
25. (New) The mouse of claim 18 wherein the DNA repair polypeptide has SEQ ID NO:2.

### Remarks

Claims 1-4, 7-15 and 19 are canceled, and claims 5-6 and 16-17 are amended, and claims 20-25 are added. The amendments are made to clarify the claims, and not for reasons relating to patentability. Therefore, the amendments are not intended to limit the scope of equivalents to which any claim element may be entitled. Claims 5-6, 16-18 and 20-25 are pending.

Amended claims 5-6 and 16-17 are supported by originally-filed claim 1 and claims 5-6 and 16-17, respectively.

New claims 20-25 are supported by originally-filed claims 2-3.

PRELIMINARY AMENDMENT

Serial Number: Unknown

Filing Date: Herewith

Title: METHODS TO ALTER LEVELS OF A DNA REPAIR PROTEIN

Page 6

D.t.: 800.019US3

The Examiner is respectfully requested to consider the amendments herein prior to taking up the application for the first Office Action.

Respectfully submitted,

JOHN H.J. PETRINI ET AL.,

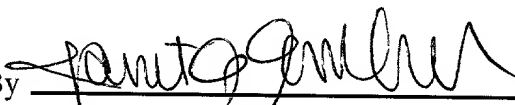
By their Representatives,

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Date

April 18, 2001

By



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This paper or fee is being deposited on the date indicated above with the United States Postal Service pursuant to 37 CFR 1.10, and is addressed to The Commissioner for Patents, Box Patent Application, Washington, D.C. 20231.

**Clean Version of Page 11, Paragraph 4****METHOD TO ALTER LEVELS OF A DNA REPAIR PROTEIN (as amended)**

Applicant: John H.J. Petrini et al.

Serial No.: Unknown

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Figure 6. Structure of the p95 cDNA. (A) The schematic diagram represents the structure of the p95 cDNA. The entire 4,483 basepair (bp) cDNA is represented by the thin line and the rectangular box is the 754 amino acid (aa) open reading frame (ORF) (SEQ ID NO:2). Within the ORF the grey box indicates the N-terminal region showing homology to *S. cerevisiae* Xrs2. The solid line above the ORF indicates the region cloned by two-hybrid screen utilizing hMre11 as bait. (B) N-terminal alignment of p95 (SEQ ID NO:3) with Xrs2 (SEQ ID NO:4). The shaded boxes indicate the regions of similarity. The two proteins show 28% identity and 46% similarity over the region displayed. The following amino acids were considered similar: {D, E, N, Q} {F, W, Y} {I, L, V} {K, R} {A, G} {S, T} {C} {H} {M} {P}. (C) A Zoo-Blot Southern blot (Clontech, Palo Alto, CA) of EcoRI digested DNA from various species was probed with the *NBS1* cDNA. Lane 1, human; lane 2, monkey; lane 3, rat; lane 4, mouse; lane 5, dog; lane 6, cow; lane 7, rabbit; lane 8, chicken; and lane 9, yeast. The position of size markers (in kilobase pairs) is indicated on the left.

**Clean Version of Page 13, Paragraph 5**

METHOD TO ALTER LEVELS OF A DNA REPAIR PROTEIN (as amended)

Applicant: John H.J. Petrini et al.

Serial No.: Unknown

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Figure 14. cDNA sequence of p95 (SEQ ID NO:1).

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**Clean Version of Page 73, Table 1**

METHOD TO ALTER LEVELS OF A DNA REPAIR PROTEIN (as amended)

Applicant: John H.J. Petrini et al.

Serial No.: Unknown

Table 1

Peptides Obtained From Mass Spectrometry Analysis

Peptide <sup>a</sup>	Position <sup>b</sup>
-QPPQIESFYPLDEPSIGSK-	189-209 (SEQ ID NO:9)
-LSSAVVFGGGEAR-	238-251 (SEQ ID NO: 10)
-WISIMDMLQR-	289-299 (SEQ ID NO: 11)
-QGLRPIPEAEIGLA VIFMTTK-	300-320 (SEQ ID NO: 12)
-TTTPGPSLSQGVSVDEK-	335-351 (SEQ ID NO: 13)
-MLSQDAPTVKE-	395-404 (SEQ ID NO: 14)
-TSSNNNSMVSNTLAK-	409-423 (SEQ ID NO: 15)
-IPNYQLSPTKLPSINK-	426-441 (SEQ ID NO: 16)
-NYFQPSTKK-	458-465 (SEQ ID NO:17)
-NKEQHLSSENEPVDTNSDNNLFTDTDLK-	503-529 (SEQ ID NO:18)
-EMDDVAIEDEVLEQLFK-	552-558 (SEQ ID NO: 19)
-MDIETNDTFSDEAVPESSK-	595-613 (SEQ ID NO:20)
-ELKEDSWAK-	625-635 (SEQ ID NO: 21)
-KLLLTEFR-	653-660 (SEQ ID NO:22)
-NPSGINDDYGQLK- <sup>c</sup>	671-683 (SEQ ID NO:23)
-EESLADDLFR-	736-745 (SEQ ID NO:24)

**Clean Version of Pending Claims**

**METHODS TO ALTER LEVELS OF A DNA REPAIR PROTEIN (as amended)**

Applicant: John H.J. Petrini et al.

Serial No.: Unknown

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5. (Amended) A method of altering the amount of a DNA repair polypeptide in a cell, comprising:
- (a) introducing into a host cell an isolated nucleic acid molecule comprising a nucleic acid segment encoding a vertebrate DNA repair polypeptide having a molecular weight of about 95000 Da as determined by SDS-PAGE, or a biologically active fragment thereof, operably linked to a promoter functional in the host cell, so as to yield a transformed host cell; and
  - (b) expressing the nucleic acid molecule in the transformed host cell as recombinant DNA repair polypeptide, wherein the amount of the recombinant polypeptide produced by the transformed cell is different than the amount of the DNA repair polypeptide produced by a corresponding untransformed cell.
6. (Amended) A method of altering the amount of a DNA repair polypeptide in a cell, comprising:
- (a) introducing into a host cell a DNA segment comprising the complement of at least a portion of a nucleic acid molecule comprising a nucleic acid segment encoding a vertebrate DNA repair polypeptide having a molecular weight of about 95000 Da as determined by SDS-PAGE, or a biologically active fragment thereof, operably linked to a promoter functional in the host cell, so as to yield a transformed host cell; and
  - (b) expressing the DNA segment in the transformed host cell as antisense RNA so as to decrease the amount of the DNA repair polypeptide in the transformed cell.

16. (Amended) A transgenic mouse whose cells contain a chimeric DNA sequence, said chimeric DNA sequence comprising:
- a transcription control sequence and an isolated nucleic acid molecule comprising a nucleic acid segment encoding a vertebrate DNA repair polypeptide having a molecular weight of about 95000 Da as determined by SDS-PAGE, or a biologically active fragment thereof, wherein the transcription control sequence and the nucleic acid molecule are operatively linked to each other and are integrated into the genome of the mouse, and wherein the nucleic acid molecule is expressed in the transgenic mouse so as to result in said mouse exhibiting increased amounts of the DNA repair polypeptide.
17. (Amended) A method of using a transgenic mouse to screen for an agent that modulates a DNA repair polypeptide, comprising:
- (a) administering the agent to the transgenic mouse, wherein the transgenic mouse comprises a chimeric DNA sequence comprising a transcription control sequence operatively linked to a nucleic acid molecule comprising a nucleic acid segment encoding a vertebrate DNA repair polypeptide having a molecular weight of about 95000 Da as determined by SDS-PAGE, or a biologically active fragment thereof, wherein the chimeric DNA sequence is integrated into the genome of the mouse, and wherein the nucleic acid molecule is expressed as the DNA repair polypeptide in the transgenic mouse; and
- (b) determining whether said agent modulates the amount of the DNA repair polypeptide in the transgenic mouse relative to a transgenic mouse of step (a) which has not been administered the agent.
20. (New) The method of claim 5, 6 or 17 wherein the nucleic acid segment comprises SEQ ID NO:1.

21. (New) The method of claim 5, 6 or 17 wherein the nucleic acid segment encodes SEQ ID NO:2.
22. (New) The method of claim 5 or 6 wherein the host cell is a mammalian host cell.
23. (New) The mouse of claim 16 wherein the nucleic acid segment comprises SEQ ID NO:1.
24. (New) The mouse of claim 16 wherein the nucleic acid segment encodes SEQ ID NO:2.
25. (New) The mouse of claim 18 wherein the DNA repair polypeptide has SEQ ID NO:2.